Circulating Microparticles and Procoagulant Activity in Elderly Patients

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Background. Microparticles (MP) are shed membrane vesicles released from activation or apoptosis of several cell types and carry a procoagulant activity. Age is associated with a procoagulant state, but the role of MP in this setting is unknown, as the relationship of MP to aging in humans. We tested the hypotheses that elderly persons compared with young persons may have different patterns of expression of MP and procoagulant activity in stable or septic conditions.

Methods. Patients from Emergency and Geriatric Departments were divided into four groups according to their age (<50 or ≥75 years old) and the presence of systemic infection (yes or no). The diagnosis of infection was reached when it was classified as certain or possible by an expert panel. Circulating MP were isolated from venous citrated blood. Cytofluorometry using specific antibodies was performed to determine the origins of MP (endothelial microparticles [EMP], red blood cell microparticles, or platelet microparticles). Procoagulant activity was determined using annexin V (prothrombinase activity) and tissue factor (TF) assays.

Results. One hundred and eleven patients were included. Elderly patients expressed a decrease in EMP in stable conditions, associated with a decrease in procoagulant annexin V MP in septic conditions (p < .05), and higher eMP levels were found in elderly infected patients who died during hospital stay than in survivors (p = .04). Compared with young patients, response to sepsis was altered in elders concerning EMP, annexin V MP, and TF-bearing MP.

Conclusion. Elderly patients expressed a different pattern of MP in stable conditions, with a different response to sepsis in procoagulant activity modification.

Key Words: Microparticles—Infection—Age effect—Elderly patients.

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AGE is associated with changes in the vasculature, hemostatic, and coagulation systems, including alterations of platelets, increased plasma levels of blood coagulation proteins, and fibrinolysis impairment (1–3). But despite the fact that age is associated with increased incidence of venous and arterial thrombotic events (1), the link between coagulation factors and risk of arterial or venous thrombosis in older populations has not been demonstrated.

Microparticles (MP) are shed membrane vesicles released from activation or apoptosis of several cell types in response to numerous stimuli (4,5). MP are bioeffectors able to modulate several biological functions, including immunity and inflammation, and carry procoagulant activity through phosphatidylserine exposure. In particular, MP represent a reservoir of blood-borne tissue factor (TF) activity, the initial activator of the extrinsic blood coagulation pathway (6–8). Whether the procoagulant activity of MP is associated with aging and that it may explain, at least in part, a higher risk of arterial events remains unknown at present.

The purpose of our study was to compare MP procoagulant activity and origins in elderly and young patients under both physiological and pathophysiological conditions. Several previous studies have reported that different circulating MP are released from platelets, endothelium, and leukocytes during sepsis (9–12) and may, at least in part, account for the occurrence of hypercoagulability (11). We made the hypothesis that age could be associated with an MP-related procoagulant state in stable or acute stress conditions, such as infection. To test this hypothesis, we measured the circulating
levels of MP from platelet, endothelial, and erythrocyte origins and measured MP procoagulant activity.

**Methods**

This study was approved by our local ethical committee (Comité de Protection des Personnes Pitié-Salpêtrière, Paris, France). According to the French law, waived informed consent was authorized because routine care of the patients was not modified.

The study was designed as a pilot study to analyze the influence of age on circulating MP levels in patients without and with infections. Other conditions known to be associated with modifications of MP levels were considered as exclusion criteria, including diabetes mellitus (13), acute coronary syndrome (14), acute ischemic stroke (15), and acute traumatic brain injury (16). Patients with statin treatment were excluded because statins are known to markedly interfere with thrombotic processes (17), including MP (18). We included patients admitted to the Geriatric Department and Emergency Department of a 1,826-bed urban teaching hospital and require a blood sampling for medical or surgical reasons. To test the effects of age and infection, patients were divided into four groups according to their age (<50 or ≥75 years old) and the presence of systemic infection (yes or no). Thus, the studied groups were as following: young controls, elderly controls, young infected patients, and elderly infected patients.

**Diagnosis of Infection**

The American College of Chest Physicians/Society of Critical Care Medicine consensus classification was used for diagnosis of systemic inflammatory response syndrome (SIRS), sepsis, and septic shock (19). All patients had SIRS defined as one or more of the following criteria: (a) body temperature higher than 38°C or lower than 36°C, (b) heart rate higher than 90 per minute, (c) hyperventilation presenting as a respiratory rate higher than 20 per minute or PaCO2 lower than 32 mmHg, and (d) white blood cell count higher than 12,000 cells/µL or lower than 4,000 cells/µL. Using clinical investigations, including body temperature, microbiological and radiological examinations, the complete medical chart, and hospitalization report, the final diagnosis of infection was determined by two independent experts in regard to the complete medical chart. Each final diagnosis was classified as certain (high probability), possible, unlikely (low probability), or absent. The final diagnosis of infection was reached when it was classified as certain or possible by the experts. In cases of disagreement among the two experts, a consensus was reached by a third expert. Experts were blinded for MP levels.

For every patient, medical care provided by the emergency physician in charge was recorded, including medical history, especially comorbid conditions, cardiovascular and neurocognitive diseases, and physical examination. All patients had blood sample for routine care management. Biological variable measurements, including hemoglobin level, leukocyte count, serum creatinine level, and C-reactive protein, were assessed in all patients with infection, in 10 of 26 young patients, and 18 of 19 elderly patients without infection. Controls of serum creatinine levels after infection treatment were obtained from hospitalization reports. According to the hospitalization report, we noticed hospital length of stay and in-hospital mortality.

**MP Isolation**

Circulating MP were isolated from 5 mL venous citrated blood within 1 hour after admission. Briefly, 5 mL of whole blood was subjected to centrifugation at 4,000g for 10 minutes to obtain plasma and then at 13,000g for 2 minutes to obtain platelet-free plasma (PFP). Then, PFP was frozen at −80°C until subsequent use. The samples were analyzed by a physician unaware of the clinical diagnosis (J.B.).

**Cytofluorometry Analysis**

Cytofluorometry was performed on PFP using a method adapted from previous validated works (20–23). Circulating levels of endothelial microparticles (EMP), red blood cell microparticles (RBC-MP), and platelet microparticles (PMP) were determined for each patient using 20 µL of PFP diluted in 80 µL filtrated phosphate buffer saline. Then, 100 µL of this solution was incubated for a period of 30 minutes with regular shaking with annexin V (100 µL of annexin V solution in the presence of 5 mM CaCl2; Roche Diagnostics, Mannheim, Germany) and/or fluorescent-conjugated antibodies, including PC5-conjugated monoclonal antibody (mAb) to CD41 (gPIIbIIIa, PMP, 10 µL; Immunootech, Marseille, France) or phycoerythrin (PE)-conjugated mAb to CD144 (VE cadherin, eMP, 20 µL) or phycoerythrin-conjugated antibodies, including PC5-conjugated monoclonal antibody (mAb) to CD41 (gPIIbIIIa, PMP, 10 µL; Immunootech, Marseille, France) or phycoerythrin (PE)-conjugated mAb to CD144 (VE cadherin, EMP, 20 µL; Beckman Coulter, Villepinte, France) or fluorescein isothiocyanate-conjugated mAb to CD235a (glycoporphin, RBC-MP, 20 µL; Beckman Coulter). A volume of 10-µm-diameter fluorescent calibrator beads (Flowcount; Beckman Coulter) equal to that of PFP plus antibodies was then added, and samples were analyzed in a Coulter EPICS XL Flow Cytometer (Beckman Coulter) operating at the medium flow rate setting. Each sample was analyzed in the same time using a fluorescent immunoglobulin G isotype in place of mAb, and this fluorescence was deduced in the calculation of MP level. MP were defined in forward scatter and side scatter modes as fluorescent events with a 0.1–1 µm diameter determined by comparison with the calibrator beads. Results are expressed as numbers per microliter of plasma. Results of the analysis were interpreted by an examiner unaware of the participant status.

**MP Procoagulant Activity**

After PFP incubation, procoagulant MP were captured by immobilized annexin V, and the anionic phospholipid
content was determined by a prothrombinase assay as previously described (24). Annexin V MP represent total circulating procoagulant MP, expressed as phosphatidylserine (PS) equivalent by reference to a standard curve constructed using liposomes of known concentrations (25). Procoagulant TF-bearing MP were captured onto insolubilized, biotinylated specific antibody to human TF and quantified using a standard TF activity assay (Innovin; Dade Behring, Marburg, Germany). Results are expressed as femtomoles of active TF.

Statistical Analysis

Data are expressed as number of patients (percentage) and mean ± SD for normally distributed variables and median and 25–75 interquartile for non-normally distributed variables (Kolmogorov–Smirnov test). Comparison of proportions was performed using the Fisher exact method. Comparison of means was performed using the Student’s t test or analysis of variance, followed by the Newman–Keuls test. Comparison of medians was performed using the Mann–Whitney test or the Kruskal–Wallis multiple comparison test. Only the following comparisons were tested: infected versus noninfected patients and elderly versus young patients. To assess the potential role of several variables that could influence the MP, a multivariate regression analysis was performed that included sex, hypertension, coronary artery disease, pulmonary infection, smoking, and atrial fibrillation. Lastly, we also performed a stepwise multivariate discriminant analysis using MP (PMP, EMP, RBC-MP, annexin V MP, and TF-bearing MP) as variables permitting to classify patients (p < .10 to be retained in the model) in the four predefined classes (young noninfected patients, elderly noninfected patients, infected young patients, and infected elderly patients). All p values were two tailed, and a p value of less than .05 was considered significant. Statistical analysis was performed using NCSS 2004 (Statistical Solutions Ltd, Cork, Ireland).

RESULTS

One hundred and eleven patients were recruited. Baseline characteristics of patients are represented in Table 1 according to their group: elderly patients without infection (n = 31), elderly patients with infection (n = 27), young patients without infection (n = 26), and young patients with infection (n = 27). For patients without infections, admission causes were mainly represented by falls (58%) and loss of

Table 1. Baseline Characteristics of Patients

<table>
<thead>
<tr>
<th>Variable</th>
<th>Young Patients</th>
<th>Elderly Patients</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control (n = 26)</td>
<td>Infection (n = 27)</td>
<td>Control (n = 31)</td>
</tr>
<tr>
<td>Age (y)</td>
<td>30 ± 8</td>
<td>36 ± 10</td>
<td>88 ± 5</td>
</tr>
<tr>
<td>Women</td>
<td>15 (58)</td>
<td>19 (70)</td>
<td>27 (87)†</td>
</tr>
<tr>
<td>Men</td>
<td>11 (42)</td>
<td>8 (30)</td>
<td>4 (12)</td>
</tr>
<tr>
<td>Previous history</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hypertension</td>
<td>0</td>
<td>1 (4)</td>
<td>14 (45)†</td>
</tr>
<tr>
<td>Dyslipidemia</td>
<td>0</td>
<td>0 (0)</td>
<td>1 (3)</td>
</tr>
<tr>
<td>Smoking</td>
<td>1 (4)</td>
<td>5 (18)</td>
<td>2 (6)</td>
</tr>
<tr>
<td>Coronary disease</td>
<td>0</td>
<td>0</td>
<td>9 (29)†</td>
</tr>
<tr>
<td>Arteritis</td>
<td>0</td>
<td>0</td>
<td>2 (6)</td>
</tr>
<tr>
<td>Stroke</td>
<td>0</td>
<td>0</td>
<td>3 (10)</td>
</tr>
<tr>
<td>Atrial fibrillation</td>
<td>0</td>
<td>0</td>
<td>3 (10)</td>
</tr>
<tr>
<td>Dementia</td>
<td>0</td>
<td>0</td>
<td>16 (52)†</td>
</tr>
<tr>
<td>Number of treatments</td>
<td>0 [0–1]</td>
<td>0 [0–1]</td>
<td>3 [2–6]†</td>
</tr>
<tr>
<td>Antiplatelets</td>
<td>0</td>
<td>0</td>
<td>13 (42)†</td>
</tr>
<tr>
<td>Infection types</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pneumonia</td>
<td>—</td>
<td>7 (26)</td>
<td>—</td>
</tr>
<tr>
<td>Urinary</td>
<td>—</td>
<td>11 (41)</td>
<td>—</td>
</tr>
<tr>
<td>Skin infections</td>
<td>—</td>
<td>6 (22)</td>
<td>—</td>
</tr>
<tr>
<td>Abdominal</td>
<td>—</td>
<td>3 (11)</td>
<td>—</td>
</tr>
<tr>
<td>Others</td>
<td>0 (0)</td>
<td>0 (0)</td>
<td>2 (7)</td>
</tr>
<tr>
<td>Death</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Biological data</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hemoglobin (g/dL)</td>
<td>13.6 ± 0.8</td>
<td>12.4 ± 1.8†</td>
<td>12.0 ± 1.6†</td>
</tr>
<tr>
<td>Leukocytes (g/L)</td>
<td>7.4 ± 1.9</td>
<td>11.2 ± 3.4</td>
<td>7.7 ± 3.5</td>
</tr>
<tr>
<td>Serum creatinine (µmol/L)</td>
<td>82 ± 17</td>
<td>72 ± 16</td>
<td>86 ± 30</td>
</tr>
<tr>
<td>C-reactive protein (mg/L)</td>
<td>19 [2–40]</td>
<td>151 [79–245]†</td>
<td>8 [4–27]</td>
</tr>
</tbody>
</table>

Notes: Other types of infection were represented by endocarditis (n = 1) and parotitis (n = 1). Results are expressed as n (%), mean ± SD, or median [25–75 interquartile]. Comparison was performed using analysis of variance and Newman–Keuls test or Kruskal–Wallis multiple comparison test. p Values refer to global comparison.

† Significant difference versus corresponding young group (p < .05).
* Significant difference versus corresponding control group (p < .05).
autonomy (16%) for elderly patients and depressive symptoms (50%) and fall injury (27%) for young patients. Types of infection were different between groups, mainly represented by pneumonia in the elderly patients (78%) and pyelonephritis in the young patients (41%). Serum creatinine level, hemoglobin level, leukocytes, and C-reactive protein were significantly different between groups (Table 1). Subsequent assessment of serum creatinine level after treatment of infection was performed, respectively, in 12 of 28 elderly patients (85 ± 22 µmol/L) and 5 of 19 young patients (70 ± 16 µmol/L) and showed no significant difference compared with the respective control groups. Seven patients died during hospital stay, all being elderly patients with infection.

Levels and types of MP were different according to age, with a significant decrease in EMP concentration but a maintenance of MP procoagulant activity (annexin V MP), PMP, and TF-bearing MP. Interestingly, infection significantly altered MP levels and procoagulant activity. Infection in young patients was associated with a significant decrease in both EMP levels and MP procoagulant activity (annexin V and TF-bearing MP). Intriguingly, this alteration in MP levels in response to sepsis was much less pronounced in elderly patients (Table 2). We found no significant difference between groups for RBC-MP and PMP. In the multivariate analysis, only hypertension (p = .02) was significantly correlated with annexin V MP, whereas the other variables (sex, coronary artery disease, smoking, atrial fibrillation, and pulmonary infection) were not. No significant correlation was noted between C-reactive protein, leukocyte count, creatinine, and any type of MP (data not shown). Comparison between the dead and survivors in infected elderly patients showed higher EMP levels in patients who died during hospital stay (Table 3).

In the multivariate discriminant analysis, the stepwise selection process identified only three types of MP, which were useful in classifying patients into the four predefined study groups: EMP (p = .10), annexin V MP (p < .001), and TF-bearing MP (p = .08). Two canonical functions were identified: the first one separating patients with infection and those without and the second one separating young and elderly patients. However, the intensity of separation was weaker for age than for infection (Figure 1), and the discriminant model identified only a relatively small proportion of variation (reduction in classification error = 25%).

**Discussion**

Our data, obtained from elderly and young patients admitted to hospital, bring new important results. First, we found reduced basal levels of circulating EMP but preserved MP procoagulant activity in elderly patients compared with young patients. Second, infection was associated with a decrease in both circulating EMP and MP procoagulant activity, which was more pronounced in young than in elderly patients. Lastly, infected patients who sustained high levels of EMP were at increased risk of in-hospital mortality than those with lower EMP levels.

**Age Effect**

Comparison of patients without infection showed a decrease in EMP level in elderly patients but maintenance of MP procoagulant activity. Age is associated with modifications of endothelium and its properties. Aging endothelium is thickened (26), and endothelium-dependent relaxation is altered (27,28). A difference in EMP level was expected because MP have been correlated with markers of endothelial dysfunction (29–31). But previous data suggested that...

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**Table 3. Circulating MP Levels for PMP, EMP, RBC-MP, and CRP According to Living Status at the End of Hospital Stay in Elderly Patients With Infection**

<table>
<thead>
<tr>
<th>MP Type</th>
<th>Control (n = 20)</th>
<th>Infection (n = 7)</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>PMP (CD41 + MP)</td>
<td>187 (95–332)</td>
<td>134 (117–289)</td>
<td>.53</td>
</tr>
<tr>
<td>EMP (CD144 + MP)</td>
<td>156 (65–266)</td>
<td>381 (129–551)</td>
<td>.04</td>
</tr>
<tr>
<td>RBC-MP (CD235a + MP)</td>
<td>647 (93–1,834)</td>
<td>337 (271–692)</td>
<td>.57</td>
</tr>
<tr>
<td>Annexin V MP</td>
<td>4.0 (1.8–5.6)</td>
<td>5.7 (3.2–11.1)</td>
<td>.30</td>
</tr>
<tr>
<td>TF activity MP</td>
<td>122 (94–202)</td>
<td>215 (60–436)</td>
<td>.55</td>
</tr>
<tr>
<td>CRP</td>
<td>122 (69–213)</td>
<td>105 (48–176)</td>
<td>.49</td>
</tr>
</tbody>
</table>

**Notes**: Results are given as numbers per microliter plasma (for PMP, RBC-MP, and EMP), PS equivalent (for annexin V MP), and TF equivalent (for TF activity MP) and milligrams per liter (for CRP). Results are given as medians [25–75 interquartile]. Comparison was performed using Mann–Whitney test. CRP = C-reactive protein; EMP = endothelial microparticles; MP = microparticles; PMP = platelet microparticles; RBC-MP = red blood cell microparticles; TF = tissue factor.

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**Table 2. Circulating MP Levels for PMP, EMP, and RBC-MP**

<table>
<thead>
<tr>
<th>MP Type</th>
<th>Young Patients (n = 26)</th>
<th>Elderly Patients (n = 27)</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>PMP (CD41 + MP)</td>
<td>179 (102–293)</td>
<td>95 (63–175)</td>
<td>.12</td>
</tr>
<tr>
<td>EMP (CD144 + MP)</td>
<td>374 (260–343)</td>
<td>222 [99–315]†</td>
<td>.02</td>
</tr>
<tr>
<td>RBC-MP (CD235a + MP)</td>
<td>290 (123–646)</td>
<td>349 [99–835]</td>
<td>.38</td>
</tr>
<tr>
<td>Annexin V MP</td>
<td>7.3 [6.4–9.5]</td>
<td>1.5 [0.7–5.2]†</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>TF activity MP</td>
<td>136 [118–200]</td>
<td>94 [78–157]†</td>
<td>.01</td>
</tr>
</tbody>
</table>

**Notes**: Results are given as numbers per microliter plasma (for PMP, RBC-MP, and EMP), PS equivalent (for annexin V MP), and TF equivalent (for TF activity MP). Results are expressed as median [25–75 interquartile]. Comparison was performed using Kruskal–Wallis multiple comparison test. p Values refer to global comparison. EMP = endothelial microparticles; MP = microparticles; PMP = platelet microparticles; RBC-MP = red blood cell microparticles; TF = tissue factor.

† Significant difference versus corresponding control group (p < .05).

* Significant difference versus corresponding young group (p < .05).
endothelial dysfunction was associated with higher, not lower, EMP levels as observed in our study. First, some conditions have been previously shown to be associated with an increase in MP level and could have influenced this result. However, we took special care to exclude patients with these conditions in both age-groups (13–15, 32, 33). Compared with young controls, elderly control patients had more frequently hypertension, stable coronary artery disease, and dementia, all conditions that have not been associated to lower MP production. Elderly controls were more frequently women, a condition associated with EMP increase (34). However, this was reported in genital active women, and postmenopausal women were not studied. In the present study, gender effect was ruled out by multivariate analysis. Finally, we think that this decrease in EMP level was associated with age per se. Basal EMP level depends on the balance between endothelium repair and stress. Age-associated decrease in EMP level could reflect a slowing of the turnover process and be an indicator of a senescent endothelium and a lower cell metabolism and activity. Interestingly, despite a decrease in EMP, we observed a maintenance of procoagulant activity using annexin V MP, PMP, and TF-bearing MP in elderly patients compared with the young patients. These results suggest that MP still carry their procoagulant activity with age, possibly due to reduced ability to maintain energy-consuming plasma membrane phospholipid asymmetry, resulting in increased exposure of the anionic procoagulant phospholipid phosphatidylserine on the external layer.

**Infection Effect**

Infection was associated with a decrease in EMP levels as well as in procoagulant activity (annexin V MP and TF-bearing MP) in young patients (41%, 80%, and 31%, respectively, $p < .05$), which was less pronounced in elderly patients (10%, not significant [NS]; 51%, $p < .05$; and 31%, NS). These results further suggest an age-related difference in MP response to sepsis. Compared with noninfected elderly patients, elderly patients with infection were more likely to be smokers and to have an increase in C-reactive protein. Tobacco effect was associated with EMP increase in secondhand exposure (35), but because we observed a decrease in EMP in elderly infected patients, it is unlikely that smoking influenced our results. This was confirmed by multivariate analysis. Chronic renal failure, a frequent comorbid condition in elderly patients, has been associated with MP (21, 36, 37), but we found no significant difference between groups according to serum creatinine levels. Lastly, nature of infections was different in infected groups, with more pneumonia in elderly patients. However, we did not observe any site infection effect in the multivariate analysis.

Influence of sepsis on EMP remains controversial. In patients with severe sepsis, EMP level was higher in infected patients compared with controls, measured as CD31 + CD42 events (38). More recently, Mostefai and colleagues (39) reported an increase of EMP level in septic shock and a protective role of circulating septic MP against vascular hyporeactivity. However, in patients with multiple organ dysfunction syndrome, sepsis was associated with a significant decrease in EMP, measured by E-selectin ($p = .003$) and anti-CD144 ($p = .063$) mAbs (40). These differences could be explained by the origin of MP, which depends on the cellular type and the mechanisms of production. Cellular activation triggers a release of E-selectin–enriched EMP acting in leukocytes rolling, whereas apoptosis is associated with CD31-enriched EMP, a constitutive marker of endothelial cells (41). The comparison of our results with these previous data is difficult because our patients were not selected on a basis of severe sepsis. We did not use any severity scale, and only one of these patients was considered as having septic shock. Eight of our elderly infected patients died during hospital stay, but our study was not designed to analyze the contribution of infection severity or comorbid conditions to MP levels.

We believe that at least two important pathophysiological pathways may lead to reduced MP levels in response to infection. First, activation of circulating and tissue-associated mononuclear cells in response to infection may increase their MP clearance ability through enhanced phagocytosis. Second, the marked increase of phospholipase activity during sepsis (42, 43) is expected to lead to phospholipid hydrolysis of MP membrane–bearing PS, the preferred substrate for sPLA2 (44), resulting in reduced MP procoagulant activity, potentially preserving some protection against a hypercoagulable state. Attenuated decrease in both EMP and annexin V MP in elderly patients compared with young patients could reflect an age-related alteration of phagocytic ability and/or phospholipase activity in response to sepsis.
MICROPARTICLES IN ELDERLY PATIENTS

Life Status

We found a significant difference in EMP according to outcome. Death was associated with an increase in EMP in elderly patients with infection. Several mechanisms can be discussed to explain this result. Persistence of higher EMP levels could reflect a more severe alteration in phagocytic clearance and be a marker of poorer evolution. This could explain results observed in previous studies showing high MP level, with more severe infections, where clearance ability could be overwhelmed by the severity of tissue damage. Mechanistically, EMP impairs endothelial-dependent vasorelaxation and NO release (21,45). Increase in EMP could alter endothelial function and contribute to alteration in tissue perfusion, an important cause of organ dysfunction and death. Nevertheless, this study was not designed to analyze mechanisms and role of MP in sepsis and prognosis of patients. Further studies will be required to clarify this role. Interest in such a clinical marker in predicting the outcome of sepsis in elderly patients is evident but has to be confirmed.

Our study has some limitations. First, due to technical limitations in their detection at the time the study was conducted, we were not able to analyze MP originating from monocytes or polymorphonuclear cells, which could have brought additional insight into role of MP in infected patients. Moreover, using PE-conjugated mAb to CD144 for EMP, we found a highly represented proportion of EMP in healthy conditions among the circulating MP. Because we did not use others’ usual endothelial markers including CD31+/CD42+ and CD62E, comparison with previous studies needs particular attention because of this methodological aspect. Second, the effect of age was analyzed in patients with comorbid conditions admitted to the Geriatric and Emergency Departments. Under such conditions, these patients cannot be considered as genuine control patients but are typical geriatric patients admitted to the hospital. Lastly, because of the relatively small sample size, the negative results concerning the possible role of variables such as sex and smoking in the multivariate analysis must be cautiously interpreted as well as MP variables not retained in the discriminant model (PMP and RBC-MP).

Conclusions

Our study brings new results about circulating levels of MP, a new vascular marker, in elderly patients. Age was shown to be associated with reduced basal levels of EMP but with preservation of MP procoagulant potential. Furthermore, age was associated with an alteration of MP response to sepsis, leading to a less pronounced reduction of EMP levels and MP procoagulant activity. Sustained elevation of EMP levels in elderly infected patients was associated with increased in-hospital mortality. This raises the question of the potential role of EMP as prognostic marker in elderly patients with infection.

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